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GROWTH AND FEEDING OF HONEYBEE LARVAE

By

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CONTENTS

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PART I. THE RATE OF GROWTH OF THE HONEYBEE LARVA.

CONTENTS.

INTRODUCTION.

Notwithstanding the abundant literature on the adult honeybee, there is a relatively small amount of exact information concerning the life history of the larva, particularly with regard to the extraordinary rate of development and the factors influencing this growth. Straus (*9*),[1] in connection with a study of the chemical composition of honeybee larvæ at different ages, gives a series of careful weighings of larvæ at successive age periods 24 hours apart.

[1] Reference is made by number (*italic*) to "Literature cited," p. 24.

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His paper, however, discusses mainly the results of metabolism in relation to age, as determined by chemical analyses of the larvæ. The observations were made under environmental conditions which were probably somewhat different from those found in this country, and, although similar, the data can be only relatively comparable to those obtained in this country. Part of Straus's data are used in Table 2 and Figure 2.

The following data and observations are presented to clear up certain points about which there is need of information and deal especially with the rate of growth of the larva, which not only is of interest from a scientific point of view but also is of value to the breeder of queen bees. Observations concerning some additional factors influencing the rate of growth of the larva, as well as information concerning the time of change in the composition of the larval food, are also given.

Part of the work herein recorded, referred to under lots 1 to 7, was done during the years 1915 and 1916 by the senior author. This early work was done independently of the work of Straus (*9*). The observations referred to under lot 8 were made mainly during August and part of September, 1922, by the junior author independently, and are supplementary to the earlier work. In connection with these observations and some made during the same summer by Mr. Lineburg, which are described by him in Part II of this bulletin, further observations were made on the amount and nature of the larval food in relation to the rate of growth. These latter were made by the junior author assisted by Mr. Lineburg.

METHODS, LOTS 1 TO 7.

Recently hatched larvæ were selected by examination, and the cells containing them were marked with a quickly-drying paint, solutions of anilin stains in alcohol being found good for such temporary use. The comb containing these larvæ was removed at fixed intervals, larvæ of known ages were removed and weighed in carefully balanced watch crystals, and in case there was any adhering larval food this was first washed off and the larva dried. While the method of choosing larvæ according to their apparent age is open to objection, it is believed that errors arising from this source are insignificant, especially in the case of larvæ chosen from new comb, as in the case of lot 1. Toward the close of the experiments another method was used which promises to give more accurate results. Newly hatched larvæ are quite uniform in size and are flexed in an arc corresponding roughly to a semicircle having a diameter of about 1 millimeter. Eyepiece micrometers were constructed having in the center of each a circle of wire with an inside diameter of 1.10 millimeters. The appearance of the newly hatched larva with respect to the wire circle is shown in Figure 1. This method, when used with a binocular microscope with extensible arm, permits an accurate selection of larvæ of minimum size.

OBSERVATIONS, LOTS 1 TO 7.

WEIGHT OF EGGS.

On July 10, 1915, 10 eggs about ready to hatch were removed and weighed, the total weight being 0.8 milligram, average 0.08 milligram.

On July 12, 20 eggs from the same colony, proving on microscopic examination to be only a few hours old, weighed 2.65 milligrams, average 0.132 milligram. On July 13, 20 eggs from the same frame weighed 2 milligrams, average 0.1 milligram. On July 27, 1916, 20 eggs taken at random, and found on examination to be in different stages of development, weighed 2.3 milligrams, average 0.115 milligram. Since newly hatched larvæ weigh about 0.1 milligram, this may be taken as the average weight of eggs about to hatch, earlier stages being somewhat heavier.

RATE OF GROWTH OF LARVÆ.

Data regarding the rate of growth were secured in 1915 and 1916 for seven lots, five of these representing weighings made at 24-hour intervals (Table 1). The histories of these lots were as follows:

Lot 1 (July 14, 1915).—Ten larvæ of fairly uniform size from a single frame, surrounded by larval jelly which covered one-third to one-half of the bottom of their respective cells, were found to weigh exactly 1 milligram, and may therefore be considered newly hatched. About 50 cells containing such larvæ, all on the same side of the frame, were marked. Weighings were thereafter made at 24-hour intervals, 10 larvæ taken at random being included in the first two weighings and 5 in each of the three succeeding. The weights of this and other lots are given in Table 1. On the third day there was evident a noticeable difference in the size of the larvæ, one being appreciably larger than the others. This individual weighed 25.3 milligrams, the average weight of the other four being only 12.75 milligrams. When seen curled up in their cells, larvæ of this age occupy from 60 to 75 per cent of the diameter of the bottom of the cells. On the fourth day the weight of the smallest of the five larvæ was 50 milligrams; that of the largest, 89.5 milligrams. The larvæ at this time filled the bottom of the cells snugly and there was no trace of larval jelly. On the fifth day the two largest in the lot of five weighed together 328.15 milligrams, an average of 164.07 milligrams. The remaining three together weighed 450 milligrams, an average of 150 milligrams. Two of the five were sealed, two not sealed, and one partly sealed.

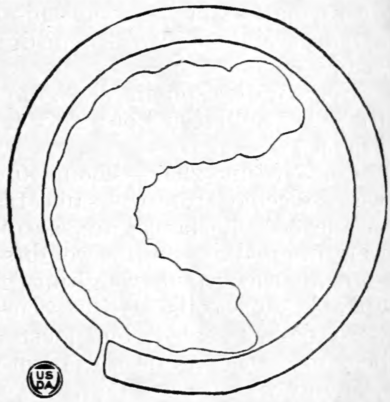

FIG. 1.—Outline sketch of newly hatched honeybee larva, as seen through the microscope in conjunction with an eyepiece micrometer composed of a fine wire ring, having an inside diameter of 1.1 millimeters.

Lot 2 (June 28, 1916).—A number of cells on a single frame containing larvæ chosen as newly hatched were marked. On June 29, 10 larvæ weighed 10.2 milligrams, or 1 milligram each, which is more than twice the weight of the larvæ of the same assumed age in Lot 1. This of course means either that the larvæ originally selected were too old or that the rate of growth during the first day was more rapid in this case. This difference continues up to the

time of sealing, so that it seems probable that these figures are somewhat too high throughout. At the second weighing (June 30) the larvæ were seen to differ greatly in size, ranging from 2.3 milligrams to 14.3 milligrams. The largest in the lot was some distance from the margin of the frame near a patch of brood about 4 days old. On July 1, larvæ 3 days old also displayed great differences in size. Three larvæ from the end of the frame, close to the brood-nest, were larger than the other two and weighed 54.4+ milligrams each. The two smaller, from the lower edge of the frame, weighed only 12.6 milligrams each. These smaller larvæ were still being fed larval jelly, while the three larger ones were being fed honey and pollen. On July 2, great differences in size were still evident, even between larvæ in adjacent cells. Two larvæ were beginning to be capped, one of these weighing 164.1 milligrams. The smallest larva in the lot weighed only 77 milligrams. On July 3, all larvæ of this lot were capped except one, which was partly capped. Capped larvæ of this and the three succeeding lots were not weighed. Weighings of 15 mature larvæ from this colony during the same season gave a total weight of 2,374.7 milligrams and an average of 158.31 milligrams.

For comparison in the table these weights are used for larvæ 4½ to 5 days old for Lots 2 to 4, and for larvæ 5½ to 6 days old for Lot 5.

Lot 3 (June 29).—About 40 cells containing newly hatched larvæ were selected from the same frame as Lot 2. At the first weighing (June 30) the larvæ appear quite uniform in size. At the second weighing (July 1) the weights ranged from 3.3 to 15.5 milligrams, and at the third weighing from 13.1 to 30.3 milligrams. On July 3, some cells of this series were completely sealed, one or two were partly sealed, but most of them were still open. Near the bottom of the frame there were three cells of this lot in a row; the two end cells were sealed, while the middle one was still open. The two sealed larvæ weighed 158.5 milligrams each; the one in the open cell weighed 117 milligrams. Another larva from an open cell weighed only 90.5 milligrams while a fifth weighed 114.3 milligrams. All the cells in this lot were sealed on July 4.

Lot 4 (July 5).—About 50 cells on the same frame were marked, great care being taken to mark only those cells containing larvæ of minimum size. On July 6, 5 larvæ weighed 3.05 milligrams, an average of 0.61 milligram. On the opposite side of the frame near a patch of brood were much larger larvæ belonging to this lot, one of the largest of these weighing 2.2 milligrams. On July 7 the larvæ in marked cells continued to show considerable differences in size. The larvæ of the group isolated among cells containing eggs remained smaller than those on the opposite side of the comb, located next to older larvæ. Most of the larvæ at this time covered a circle one-half to two-thirds of the diameter of the bottom of the cell. The total weight of five larvæ taken at random from both sides of the comb was 31.75 milligrams, an average of 6.35 milligrams. The individual weights, in milligrams, were as follows: 2.85, 3.7, 3.7, 9.55, 11.95. All larvæ were still fed with larval jelly. On July 8, the weights of 5 larvæ ranged from 17.5 to 55.3 milligrams. The larger were uniformily near unsealed brood, the smaller among eggs near the middle of the frame. The smaller larvæ occupied less than

two-thirds of the diameter of the bottom of the cells; the larger snugly filled the bases of the cells. On July 9 the weight of 5 larvæ was 502.7 milligrams or 100.54 milligrams each, the larvæ near brood continuing to exceed in size those on the opposite side of the frame. The mouths of the cells containing the two largest larvæ in this lot were beginning to be contracted. The weight of these two larvæ was 284.9 milligrams or 142.45 milligrams each. The two smallest larvæ in this weighing weighed 121.5 milligrams, or 60.75 milligrams each.

Lot 5 (July 6).—A new comb in the same colony was found to be full of eggs and young brood. About 60 cells on one side of the comb were marked, only the very youngest and most minute larvæ being chosen. Some of the larvæ were so recently hatched that they had not yet been supplied with food. The first weighing of 10 larvæ at the age of 1 day showed an average of 0.52 milligram. This is lower than the average weight for the same (estimated) age of any of the previous lots, except Lot 1. None of these larvæ were near older unsealed brood. On July 8 the average weight, 2.96 milligrams, approximated that of Lot 1 of the corresponding age, the individual weights ranging from 2.12 to 3.75 milligrams. On July 9 considerable differences in size appeared. The average, 14 milligrams, is close to that for the corresponding age in Lot 1, but a considerable range in size became evident—8.75 to 22.15 milligrams. The larvæ at this time covered a circle whose diameter is one-half to three-fourths that of the bottom of the cell. On July 10, the total weight of 5 larvæ was 295.2 milligrams, the individual weights ranging from 41.8 to 78.5 milligrams. On July 10 about 15 marked cells which contained larvæ remained; 3 of these were completely sealed, 2 one-half sealed, and 4 just beginning to be sealed. Weights were as follows:

	Milligrams.
1 larva completely sealed	171.3
1 larva one-half sealed	154.5
1 larva less than one-half sealed	151.7
1 larva with edge of cell slightly turned in	126.8
1 larva with edge of cell slightly turned in	128.6
Total weight	732.9
Average weight	146.6

Lot 6.—Fifteen to twenty larvæ from the same frame were selected as follows: The end of a wire 1 millimeter in diameter was ground off square, to be used as a measure, and only those larvæ were selected which in their normal position in the cell could be completely covered by the end of the wire. Such larvæ were approximately newly hatched. On the third day, larvæ were found in only five marked cells. These averaged 26.1 milligrams. The individual weights in milligrams were as follows: 14, 17.8, 19.3, 36.9, 42.5.

Lot 7.—On July 20 at 9.15 a. m., the queen in the same colony was placed in the center of the hive on a frame free from brood in a space partitioned off from the remainder of the colony by queen-excluding zinc. At 11.15 a. m. some eggs were found on the frame and the queen was removed, great care being taken to prevent her from returning to this comb. The hive was then closed and left undisturbed for three days. On July 23, at 3 p. m., this comb was examined under a binocular microscope and several larvæ were seen

surrounded by food. All seemed large as measured by the eyepiece micrometer. One unhatched egg was seen. According to previous observations of the writer, these eggs should have hatched between 11.15 a. m. and 1.15 p. m. on this date. On July 26, 5 of the 13 larvæ present were weighed, the individual weights in milligrams being as followed: 5.05, 6.1, 20.9, 23.4, 28.9. Considerable differences in the size of the larvæ were evident on examination of the comb. Three of the larvæ covered an area of less than one-half of the diameter of the bottoms of their respective cells, but most of them nearly or quite filled the bottoms of the cells. All of the larvæ were supplied with an excess of larval food.

VARIATIONS IN RATE OF GROWTH.

An outstanding feature of these observations is the great individual variation in the daily rate of growth as manifested in the differences between the weights of the 5 to 10 larvæ of the same age in different lots, as well as in the differences between individuals of the same age in the same lot. These considerable differences naturally led to the suspicion that they might have been due to the method used in selecting newly hatched larvæ. The experiment described under Lot 7 was intended to test this supposition. As already stated, larvæ 3 days old, hatched from eggs laid within a determined 3-hour period, showed wide divergences in individual weight. There were on hand two series of larvæ, each from a single batch of eggs laid within a known period of 2 hours, preserved in alcohol. The greater part of these were fixed in their cells, the earlier stages being embedded in the coagulated larval food and therefore unsuitable for weighing. Five larvæ 3 days old of one lot were free from foreign material and suitable for weighing for comparison with one another. These showed on superficial examination considerable differences in size. These larvæ were weighed separately, the weights in milligrams being as follows: 5.7, 7.35, 7.5, 9.8, 10.4. Although the differences in weight between the different individuals of this lot are not as great as between those of most of the other lots, still they are considerable, the difference between the smallest and the largest amounting to nearly 100 per cent. This result, taken in conjunction with the great differences between larvæ of the same age in different lots, makes it reasonable to conclude that considerable differences exist in the rate of growth of bee larvæ.

In fly larvæ Herms (*2*) determined that "there is an optimum when enough nourishment has been taken to pass through the metamorphosis to the best advantage." This optimum is evidently represented in the honeybee by a weight of about 158.31 milligrams when the larvæ are sealed by the worker bees. As far as the preceding observations on the honeybee are concerned, the optimum weight virtually coincides with the average weight at maturity.

LENGTH OF THE LARVAL PERIOD.

As the data show, sealing may begin as early as the end of the fourth day after hatching (Lots 2 and 3) and as late as the middle of the fifth day (Lots 1 and 5), the average for the five lots being about 4½ days (Table 1). It is noteworthy that a difference of half a day may exist between two lots from the same hive, not only during

the same season but with approximately the same weather conditions. In the case of two earlier lots from eggs of known age, not here recorded in detail, the larvæ were sealed between 5 and 5½ days, the sealing beginning when the larvæ were about 5 days old and continuing for some hours.

The individual differences in the time of sealing are well illustrated by the larvæ of one of these lots. These were all of equal age (within 2 hours), located on the same comb and therefore presumably subject to approximately identical environmental conditions. When the larvæ were approximately 5 days old, there were 43 larvæ on one side of the comb. Six of these were already sealed, while about half of the remainder showed a thin rim of wax around the mouths of the cells, indicating that capping had just commenced. Two and a quarter hours later about one-fourth of those cells open previously were sealed. At this rate it would require from 8 to 10 hours additional for the sealing of all cells to be completed. Such differences are typical of all the lots.

The data given for Lot 5 suggested that the sealing of the cell is commenced before the larva has attained maturity. To test this conclusion additional weighings were made from the same colony as follows:

	Milligrams.
4 larvæ from cells whose edges were just beginning to show signs of capping averaged	137.60
5 larvæ in which sealing was slightly more advanced averaged	142.20
10 larvæ from cells one-fourth to one-third sealed averaged	141.00
10 larvæ from cells one-third to one-half sealed averaged	158.21
10 larvæ from cells over one-half sealed averaged	157.20
15 larvæ from freshly sealed cells averaged	158.31

These figures show clearly that the sealing of the cell is actually commenced when the larva lacks about 20 milligrams of its final weight, and that the larva is fed until the cap covers about one-third of the mouth of the cell, at which time the larva attains its maximum weight. The completion of the cap probably occupies only a short space of time. The weight of mature larvæ is also shown to be quite constant, at least for a given colony at the same season of the year. Only one marked divergence from the average for this colony was noted, this occurring in Lot 5, in which a single sealed larva was found to weigh 171.3 milligrams.

EFFECTS OF UNDERFEEDING.

It has been shown by Herms (*2*) and Whiting (*11*) for two species of the fly Lucilia (*L. caesar* L. and *L. sericata* Meig.) that undersized flies are produced by underfed larvæ. A similar condition is found in the boll weevil (*Anthonomus grandis* Boh.), in which it is stated that the size of the adult varies in almost direct proportion to the abundance of the larval food supply and the length of the period of larval development (*3*).

The following experiment was tried to determine whether the same relation exists between larvæ and adults of the honeybee. Ten larvæ, all decidedly below the average maximum weight, and therefore not fully fed, were taken from a normal colony, weighed and placed in artificial cells formed by making several deep cylindrical

grooves one-fourth of an inch in diameter in the surface of a flat block of wood which had been impregnated with melted wax. These cells were closed at the ends with wax and at the top with cover slips. The larvæ were then placed in an incubator and kept at 37.5° C. All of these larvæ spun more or less perfect cocoons, but only two succeeded in forming normal pupæ. From these two pupæ perfect adult bees emerged. The weight of these in milligrams, as compared with their larvæ and the weights of normal larvæ and pupæ, are given below:

	Larva.	Adult.
No. 1	115.0	68.1
No. 2	144.1	94.2
Normal	158.3	112.0

Since adult bees from fully fed larvæ reared under similar artificial conditions are only slightly under normal weight, it is possible to produce undersized bees from larvæ that are not fully fed. Undersized or dwarf bees are familiar to most beekeepers, but whether they are due to underfeeding of the larvæ is not definitely known.

METHODS, LOT 8.

In order to counteract variations observed in the earlier experiments as much as possible, an attempt was made during the work of 1922 to perfect the method suggested under Lot 7, which seemed to give promise of the best results. A space in the middle of a hive wide enough for two brood frames was partitioned off with queen-excluding zinc at either side and at the entrance. This was to prevent the queen from escaping in any way after the cover was placed on the hive, but the workers could come and go at will. A fairly strong colony of bees was transferred to this hive, placing one frame nearly filled with brood in this partitioned-off space, with a second frame either empty or with only a small amount of sealed brood in it. The queen was then confined in this chamber. The nearness of the well-filled brood comb to the empty comb generally induced the queen to lay a fairly large number of eggs in a period of 12 hours or less.

To obtain average live weights of larvæ of known age at definite age periods frames of eggs were removed from this chamber after a definite length of time. The frame removed was replaced by another empty one for the next series. The removed frame of eggs then was kept with several other frames of brood covered with nurse bees in a super over a strong colony, with the queen confined below by a queen excluder. After 3 days' incubation as eggs, at intervals of 24 hours each, larvæ were removed by means of small forceps for weighing. So far as possible, the time of day selected for weighing the larvæ corresponded to the middle of the egg-laying period for that series. A method of weighing similar to that previously employed was used, except that, to reduce the probable error, five larvæ were taken for each weighing instead of individual larvæ. Larvæ of average maximum size for the age period, and as nearly alike in size as possible when selecting from gross appearances, were chosen for each group of five. These were carefully removed, cleaned of food material, placed in a watch crystal, and weighed immediately by means of a chemical balance. Even with this method more or less variation in weight for each age period was

observed, but by comparing the weights given in Table 2 with the results given in Table 1 it may be seen that there is less variation with the method here described.

OBSERVATIONS, LOT 8.

EFFECT OF NO HONEY-FLOW.

As will be noted from Table 2, a majority of the weighings here recorded were made during August. In the region of the Bee Culture Laboratory (Somerset, Md., near Washington, D. C.) there is generally at this time of year an absolute dearth of incoming nectar and a consequent depletion of the reserve stores of the colonies. It was noticed that the average weights of the older larvæ, particularly of those over 4 days old, were consistently less than the values given in Table 1, Lots 1 to 5, for these ages, in certain cases as much as 6 to 8 per cent. In fact, many of the older larvæ that were presumably of the desired age were discarded because they were abnormally under weight. The weights for the age period below 4 days did not show this marked divergence, presumably because by the method used the variation in feeding in relation to the position of the larvæ on the frame was to a large extent eliminated. Most of the earlier observations (Lots 1 to 7) were made during June and July, at a time when there is generally some nectar flow from clover and other flowers sufficient to keep the bees from drawing on their reserve stores. It was suspected in these experiments that the lack of stimulation might be affecting the weights and rate of growth, thereby somewhat lengthening the larval period and lowering the maximum weights. The fact that the majority of the larvæ observed usually were not capped until about 18 to 24 hours later than most of those observed in Lots 1 to 7 seems to bear out this assumption of slower development.

No observations were made on the time period of emergence as adults, although the decrease in weight continued into the pupal period. It is known, however, that with certain other insects a scarcity of food often materially lengthens the developmental period. As stated above, insufficient food has a marked effect upon the weights of larvæ and even of adults.

Honeybee larvæ are peculiarly adapted for rapidly consuming and assimilating large amounts of food. Little energy need be expended except for the slight movements of feeding. Therefore practically all the food assimilated is used for the growth of tissues or is stored for future use as required during pupation. As a result the rate of growth should be more or less proportional to the composition and amount of the food consumed, and probably would be more noticeable with the more simple food of the later stages. Anything affecting the amount of this unpredigested food would consequently affect the rate of growth.

EFFECT OF STIMULATION.

In an attempt to demonstrate whether the above assumption is correct, several series of larvæ were subjected to conditions more nearly resembling the stimulative effects of a honey-flow. Frames containing eggs of known age were placed for development in a

strong colony which was fed a thin sugar sirup throughout the experiment, in order to stimulate activity. As a result, as will be noted from Table 2 and Figure 2, the weights of the older larvæ were noticeably increased under these conditions. In the case of the maximum-sized larvæ at the 6-day age, this amounted to as much as 8.1 per cent. This series was on the average sealed earlier than the previous series. These figures also more nearly correspond with those of Table 1.

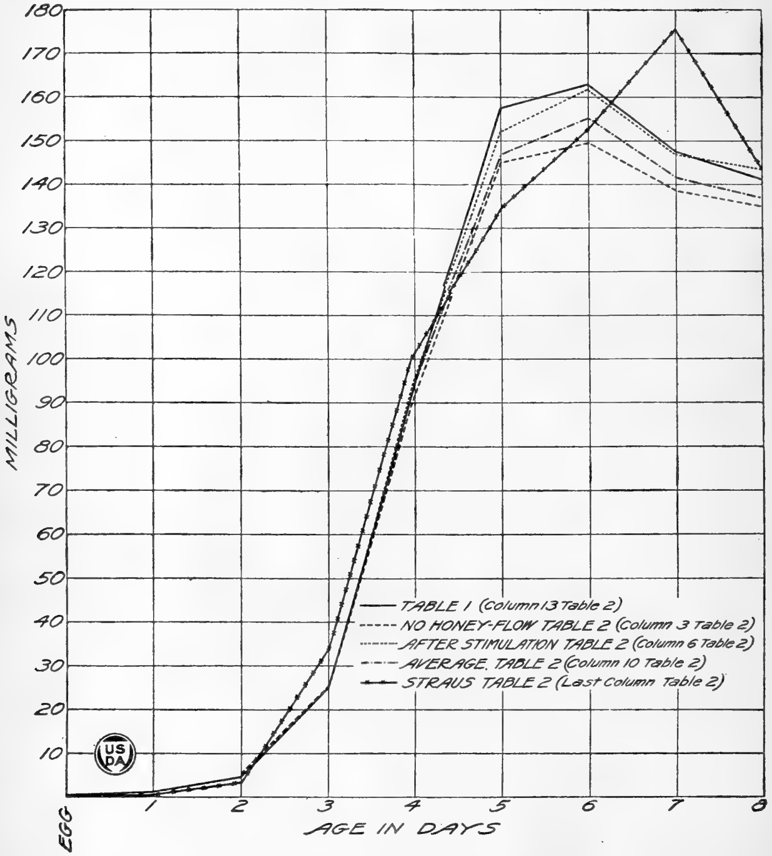

FIG. 2.—Living weights of honeybee larvæ at different age periods and under varying conditions. (Tables 1 and 2.)

Therefore, it seems safe to conclude that the amount of incoming nectar has an appreciable effect upon the development of at least the older larvæ. Weights of larvæ under 3 days old did not show this variation to any noticeable degree, there being only 1.7 per cent difference for the 4-day age and less for still younger ones. The curves shown in Figure 2 illustrate this point graphically. It will be noticed that the various growth curves, even to a relative extent that drawn from the Straus data, are almost identical for the first three days, only beginning to diverge on the fourth day.

NATURE AND COMPOSITION OF BROOD FOOD.

The foregoing observations have a bearing on the much discussed question of the source and nature of brood food at different times during the feeding period. The relation of the composition and amount of brood food to the rate of development may assist in explaining the variations observed above.

To obtain the weight of food unconsumed in the cells, as an indicator of the amount of food consumed in relation to the weight of larvæ of approximate known age, the following method was used:

A frame of brood was chosen from a normal colony, containing in an area of 2 or 3 square inches several larvæ of uniform size. The age of these larvæ was approximately determined by inspection and comparison in size with the drawings of larvæ of definite age (figs. 6 to 11). A piece of this comb was then cut out containing about the desired number of larvæ. The edges of this piece of comb were trimmed clean with a sharp scalpel and the cells of the comb were cut down so as to reduce the depth of the cells to about half. The edges and all cells containing larvæ of abnormal size not desired for weighing were cleaned and dried with swabs of absorbent cotton on the ends of matches. This was done as rapidly as possible so as to prevent loss by evaporation from the remaining cells. The piece of comb was then carefully weighed. The larvæ were then removed with fine forceps, care being taken to remove as little adhering food material as possible, and then the piece was weighed again. The difference between these two weights gives the approximate weight of the larvæ (Table 3). To obtain the weight of the remaining food, each cell was cleaned with a swab of absorbent cotton and the cell was then dried out thoroughly with a second swab to insure complete removal of all the food material. The weight of the empty comb was taken, by which the weight of the food material is determined. The average age of each lot was more approximately determined by comparison and interpolation with the weights of larvæ of known age, given in Table 2.

Unfortunately, owing to lack of time and the small number of determinations made, the data given in Table 3 are only approximately accurate. The data are suggestive, however, and considerable interesting information may be obtained by correlation with the rate of growth, the time of change in food composition, and the time spent by nurse bees in caring for the larvæ of various ages.

It is noticeable that the small, young larvæ are always surrounded by or even are floating on an excessive amount of food material, which has a uniformly grayish-white, pastelike consistency. From the data observed (Table 3, figs. 3 and 4), it was found that up to between the second and third days this excess of food, as indicated by the amount unconsumed in the cell, is considerably greater than the weight of the larva itself. There has been much controversy as to whether the food of the young larva is a secretion from glands or a regurgitation from the ventriculus [Snodgrass (8)]. Its consistent lack of pollen grains and uniform appearance suggest secretion rather than regurgitation, since if regurgitated it would of necessity contain undigested pollen grains, as these are always found in the ventriculus. If a secretion, the food of the younger larvæ would be affected much less by variations in external conditions, such

as dearth of nectar, while the food consisting mainly of honey and pollen, such as is fed to the older larvæ, could be affected by such circumstances, as the data given suggest.

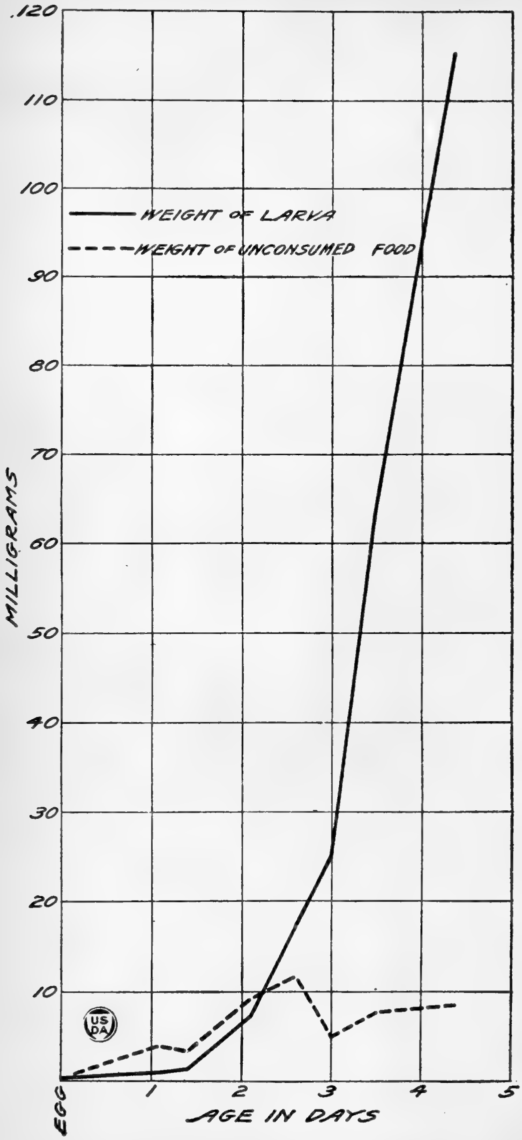

FIG. 3.—Weight of unconsumed food in the cell in relation to weight of honeybee larva. (Table 3.)

TIME OF CHANGE IN COMPOSITION OF BROOD FOOD.

It has been shown by the investigations of von Planta (7) (Table 4), that the younger larvæ receive a food that is much richer in fat and albuminous constituents than that given the older larvæ. In the case of the older worker larvæ this food is discontinued and a food consisting of a mixture of nectar (or honey) and pollen is substituted. The latter food was shown to be much higher in sugar content, while the fat and albuminous content decreases considerably. This food also contains undigested pollen grains, which are absent in the food of the younger larvæ. Von Planta stated that this change in the composition of the food occurs when the larva is 4 days old. From the observations of the writers, as well as of others, however, this change is found to take place as early as during the third day. This was demonstrated by the fact that throughout these experiments undigested pollen grains were regularly found in the food of larvæ no more than 3 days old from the time of hatching from the egg, often in sufficient amounts to color the food slightly. Also, as mentioned above, the coincidence of the curves in Figure 2 for rates of growth up through the third day even under

varying conditions strengthens this assumption. Larvæ destined to become queens, however, receive a highly nutritious food, "royal jelly," produced by the worker bees, throughout the entire larval period. It is well known that queen larvæ at maturity are larger and heavier than worker larvæ. Unfortunately, no weighings of queen larvæ were made, so that figures are not available for comparison with those of worker larvæ, but it is safe to assume that although they would raise somewhat the percentage increases for the last two days, yet the drop would doubtless still be evident.

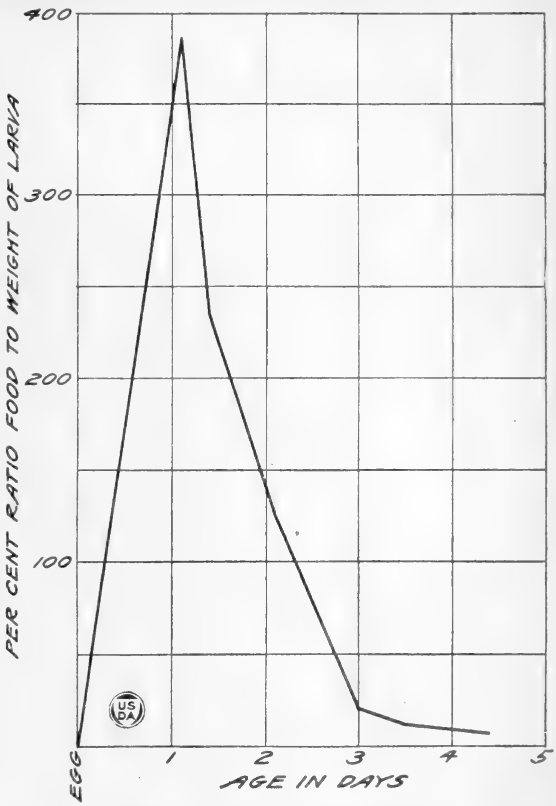

FIG. 4.—Per cent ratio of unconsumed food in the cell to weight of honeybee larva. (Table 3.)

RATE OF GROWTH.

The weighings recorded above are too few in number to justify broad generalizations, but are nevertheless of value in some particulars. The most obvious and striking feature is the remarkably rapid growth of the larva, which within 4½ to 5 days increases its initial weight more than 1,500 times. The ratio of initial weight to weight at maturity of several insects is shown below:

Cossus (*4*)	1	: 72,000	(Lyonet).
Sphinx (*6*)	1	: 9,976	(Newport).
Bombyx (*1*)	1	: 9,500	(Dandolo).
Telea (*10*)	1	: 8,600	(Trouvelot).
Apis	1	: 1,580	
Anthophora (*6*)	1	: 1,020	(Newport).
Sarcophaga (*2*)	1	: 451	(Herms).
Lucilia (*2*)	1	: 404	(Herms).

The first four of the insects listed above, as compared with the honeybee, have an extended larval period—3 years in the case of Cossus—and their food is of low nutritive value. In the case of the last four, particularly the honeybee, the larval food is of high nutritive value and is rapidly assimilated. In rate of growth the honeybee larva exceeds all of those listed, followed by the fly Sarcophaga, which attains 451 times its initial weight in about 71 hours.

In larvæ of the flies Sarcophaga and Lucilia, Herms (*2*), using the percentage increment method of Minot (*5*), found that "the tendency is for growth to decrease in rate uniformly." This rule does not apply to the honeybee, however, as the averages and percentage increases of all lots show (Tables 1 and 2). Moreover, the average per cent in each lot tends to reach a maximum at the end of the second day, with the exception of Lot 2 (Table 1). The individuals of this lot are open to the suspicion of being actually older than estimated. This suspicion, if confirmed, would explain the circumstance of the maximum per cent increment falling at the end of the first instead of at the end of the second day. After the second day the average percentage increment of the five lots combined shows a steady drop until maturity.

CORRELATION OF FOOD WITH THE RATE OF GROWTH.

Physiologically food may be defined as any substance taken in by an organism and made use of for any purpose. Nitrogenous material is necessary for growth and repair of tissues, no large amount being stored, except as tissue or protoplasmic substance. Sugars and fats are readily assimilated and stored in the tissues as glycogen and neutral fats and are sources of energy by oxidation for various purposes.

It is seen from Figure 5 that the greatest per cent of daily increase in growth for the honeybee larva takes place during the first three days of larval life, with the maximum at two days. After three days the per cent of daily increase is noticeably less, although the actual increase in weight of the larva rises rapidly to the maximum just after sealing has taken place. According to results of the analyses by Straus, when, during the first three days, the nitrogenous content of the food is high for the purpose of tissue building, relatively little storage of glycogen or fat takes place. Since little energy is required for the slight movements made by the larva, after the change in the composition of the food takes place and it becomes predominantly carbohydrate (that is, high in sugar content from the honey used), the great storage of glycogen takes place which is to serve as a source of energy during metamorphosis. This again seems to divide the larval period at about the 3-day age.

Again, as may be observed from the data given in Table 3 (figs. 3 and 4), at the time when the greatest relative increase in weight occurs and the food has the highest growth-producing, nitrogenous content, there is a marked excess of unconsumed food in the cells nearly four times the weight of the larva 1 day old. It will be noted that the weight of the 1-day-old larva (0.65 milligram) plus the weight of the food unconsumed in the cell on the first day (3.96 milligrams) practically equals the weight of the larva on the second day (4.745 milligrams) (Table 2). This relation does not continue, because the excess of food decreases, and the change in composition occurs soon afterwards. After the third day the larva has developed to a size sufficient to consume an increasingly larger volume of food as soon as it is placed in the cell, particularly as probably considerable food is actually placed directly in the mouth of the larva. This accounts for the small amount or almost total absence of food in the cells for the late stages just prior to sealing. There is a slight increase in weight, however, after sealing takes place. Therefore, correspond-

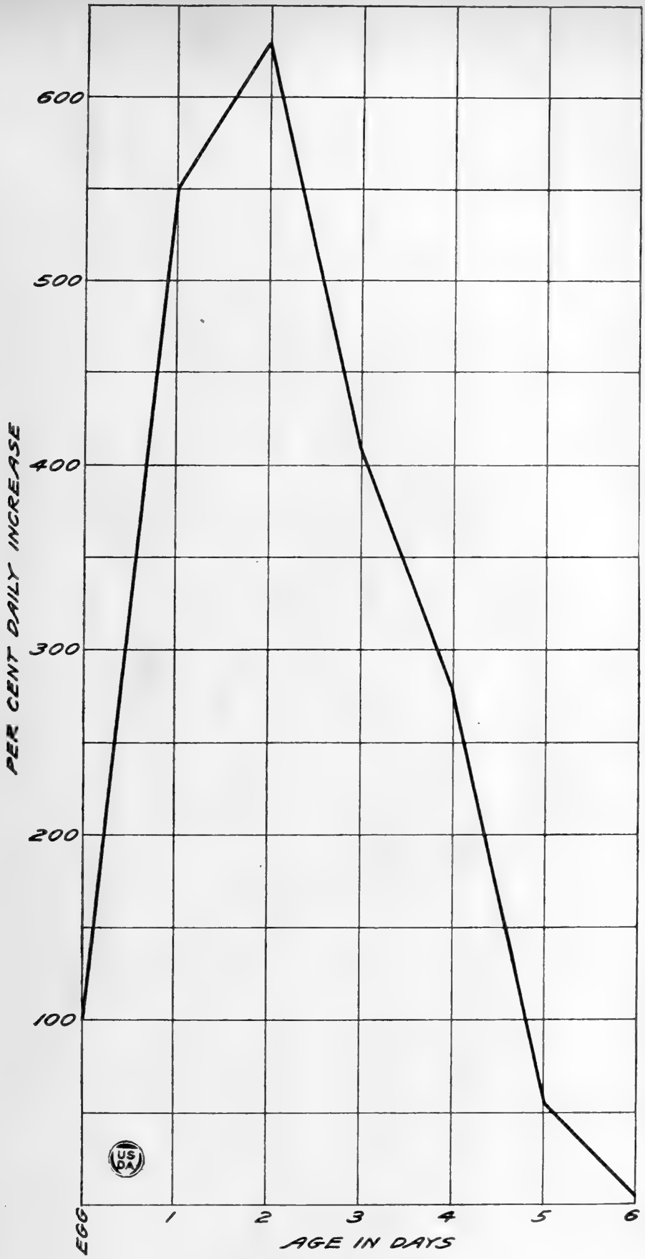

FIG. 5.—Per cent daily increase in weight of honeybee larva over weight of the preceding day. (Table 2.)

ing with the time of change in composition, the excess of food unconsumed in the cells greatly decreases in relation to the weight of the larva (fig. 4).

CORRELATION WITH TIME SPENT IN NURSING.

The effect of these food requirements upon the amount of work necessary to be done by the nurse bees in relation to the rate of growth of the larvæ and nature of the food is well illustrated by the observations of Lineburg in Part II of this bulletin. He found that in the time spent by nurse bees in caring for larvæ there is practically 100 per cent increase on the first day over that at the time of hatching from the egg. Food is never placed in the cell until soon after the larva hatches. On the second day this again decreases about an equal amount, but begins to increase again to a remarkable degree, particularly after the third day. From the third day on the living weight (fig. 2) and the time spent in feeding (fig. 13) show a remarkable correlation. The high point for feeding on the first day corresponds with the maximum excess of unconsumed food in the cells at this time. It seems probable that the nurse bees place an excess of the predigested food in the cell soon after the egg hatches, sufficient for the larva for about two days, so that only a minimum of attention is required until the change in the composition of the food takes place, at which time increasingly greater demands are made by the larva for honey and pollen.

FIG. 6.—Larva of honeybee newly hatched from the egg.

Figures 6 to 11 were all drawn with the aid of the camera lucida from material preserved in alcohol. A special effort was made to secure as far as possible specimens representative of the average size of larvæ of the age designated. In figures 6 to 10, inclusive, the larvæ are represented as seen from the mouth of the cell, the outlines of the latter being represented by a hexagon and drawn accurately to scale.

Since the greatest relative increase in growth takes place during the period of uniform, highly nitrogenous food and the greatest storage of reserve energy-producing materials occurs after the food has changed in composition it may be assumed from the observations cited that any variations in honey-flow will affect the rate of growth of the older larvæ more than the younger larvæ.

GENERAL APPEARANCE OF LARVÆ OF DIFFERENT AGES.

The general appearance of larvæ of different ages, particularly with reference to their size and position in the cell, provides criteria by which to judge the age of the brood.

The newly hatched larva (fig. 6) has a total length of 1.6 millimeters. It is usually bent in a curve approximating a semicircle and lies on its side on the bottom of the cell. At this stage it is slender as compared with the later stages, tapers gradually from the head, and is nearly transparent. It usually lies near the center of the base of the cell in a mass of transparent larval food which does not exceed in diameter one-half that of the base of the cell. A circle drawn around the larva in its usual flexed position would closely approximate 1 millimeter in diameter (fig. 1). A larva may lie on either

its right or its left side, but in the accompanying figures the larvæ are all shown lying on the right side. Owing to the transparency of both the larva and the larval food in which it lies, it is often difficult to detect the presence of larvæ of this age without the aid of the microscope, especially when the brood comb is old and dark. In

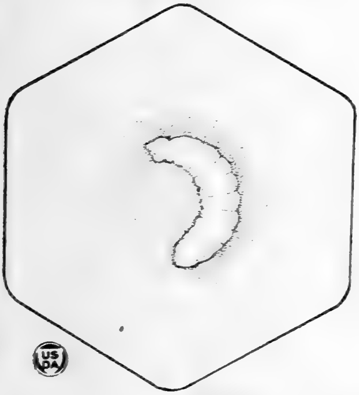

FIG. 7.—Larva of honeybee, 1 day old.

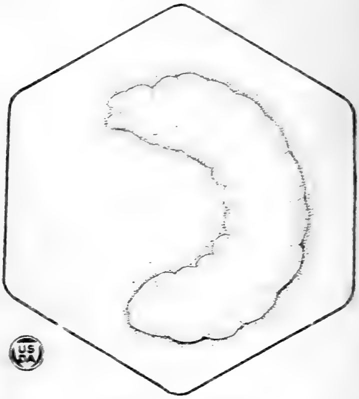

FIG. 8.—Larva of honeybee, 2 days old.

several instances newly hatched larvæ were found which were not provided with food, so that the presence of larval jelly is not necessary for the escape of the larva from the eggshell. Movement is frequently observed in young larvæ, but this is confined to simple extension and flexion of the body.

At the end of the first day the larva has attained a length of about 2.6 millimeters (fig. 7). It is now somewhat less transparent and the diameter of the posterior end of the body is now noticeably larger than that of the anterior end. The area of the base of the cell covered by larval food is proportionately greater.

At the close of the second day (fig. 8) the larva measures slightly less than 6 millimeters in length and is still surrounded by food.

Larvæ 3 days old (fig. 9) are no longer bent in a semicircle, but form an incomplete ring, nearly covering the bottom of the cell. They are much less transparent than at earlier stages and begin to assume the opaque ivory-white appearance characteristic of the later stages.

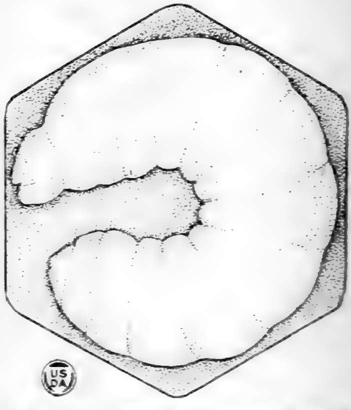

FIG. 9.—Larva of honeybee, 3 days old.

At the end of the fourth day the larva (fig. 10) is bent to form a complete ring, the anterior and posterior ends overlapping, with the anterior end always above or outside the posterior end. The larva now snugly fits the bottom of the cell, but the two ends of the larva are still plainly distinguishable. Larvæ of this age no longer have

their food surrounding them, but a small amount of honey (nectar) mixed with pollen may be found on the base of the cell and adhering to the larva.

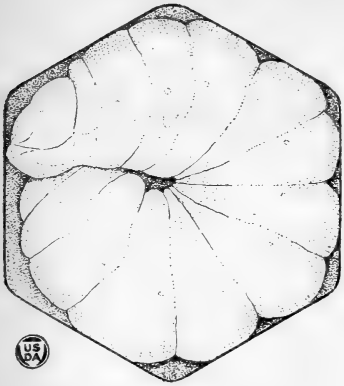

FIG. 10.—Larva of honeybee, 4 days old.

Full-grown larvæ (fig. 11)—that is, larvæ in cells more than half covered by the cap—fill the lower half of the cell so completely that the form of the larva suffers considerable distortion. The body is flattened and the head and tenth trunk segments are crowded back, telescoped into the neighboring segments, and, as the figure shows, are invisible from the mouth of the cell. Larvæ of this age, fixed and hardened in the cell, as was the one from which the figure was drawn, form veritable casts of the interior of the lower half of the cell. It should be noted that the head is always nearer the mouth of the cell than is the posterior end, the long axis of the larva thus following a spiral course.

SUMMARY AND CONCLUSIONS.

Eggs of the honeybee at the commencement of development weigh about 0.132 milligram each. At the close of embryonic development they weigh from 0.08 to 0.1 milligram each.

Larval development, up to the time of sealing, lasts $4\frac{1}{2}$ to $5\frac{1}{2}$ days. The average weights of the larva obtained from Lots 1 to 5 were as follows: First day, 0.65 milligram; second day, 4.687 milligrams; third day, 24.64 milligrams; fourth day, 94.692 milligrams; fifth day, 157.642 milligrams. Averaging the weights obtained under various conditions under Lot 8 the following weights were obtained: Second day, 4.745 milligrams; third day, 24.626 milligrams; fourth day, 93.99 milligrams; fifth day, 146.748 milligrams; sixth day, 155.005 milligrams; seventh day, 141.647 milligrams; eighth day, 137.165 milligrams; ninth day, 133.152 milligrams. Weights after the fifth day are of sealed brood.

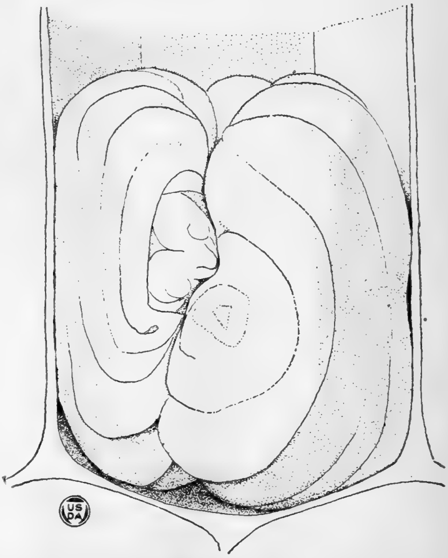

FIG. 11.—Mature honeybee larva, the half of the cell facing the observer being removed so that the larva is shown as seen from one side of the cell, and not from the outer end, the base of the cell being toward the bottom of the page.

The increase in growth, measured by percentages, tends to attain a maximum at the close of the second day and from that time steadily declines until the close of larval development.

Great individual differences in weight are noted between different lots of the same age and between different individuals of similar age in the same lot.

The maximum average weight appears to be fairly constant, at least for the same colony under similar conditions.

Stimulating the activity of the bees by feeding a thin sugar sirup during a dearth to produce the effect of a honey-flow was found to cause an increase in the weight of larvæ 4 days old and above, to as much as 8 per cent in the case of 6-day-old larvæ (just sealed).

The weights of larvæ over 3 days old are found to be affected by variations in the honey-flow, the rate of growth decreasing under conditions of dearth.

Sealing of the cell is begun before the larva has attained its final weight, and about one-third of the mouth of the cell is covered before this weight is attained.

Great differences exist in the time of the sealing of the cells of a given lot of larvæ of the same age, a period of several hours intervening between the sealing of the earliest and that of the latest cell.

Undersized or dwarf bees may be produced from larvæ that are not fully fed.

From the various observations made, the change in the composition of the food of the honeybee larva, from one of high nitrogenous content to one of high sugar content, takes place as early as the third day after hatching from the egg.

The excess of weight of food unconsumed in the cell before this change in food composition occurs is correlated with the uniformly great relative increase in growth of the larva during the first three days of larval life. All later changes in the rate of growth are correlated with the condition of the honey-flow.

The time spent by the nurse bees in nursing, as observed by Lineburg, is correlated with the demands of the larvæ for the different types of food before and after change in composition occurs.

TABLE 1.—*Weights of the bee larvæ of five lots, at intervals of 24 hours (1915 and 1916).*

Lot No. and age in days.	Total weight.	Number of larvæ.	Average weight.	Average daily increase.	Daily increase.	Remarks.
Lot 1:	*Mg.*		*Mg.*	*Mg.*	*Per cent.*	
Eggs	2.00	20	0.100			
0 day	1.00	10	.100			
1 day	4.45	10	.445	0.345	345	
2 days	27.55	10	2.755	2.310	519	
3 days	76.30	5	15.260	12.505	454	
4 days	333.10	5	66.620	51.360	337	
5 days	778.15	5	155.630	89.010	134	Larvæ partly sealed.
Lot 2:						
1 day	10.20	10	1.020	[1] 0.920	[1] 920	
2 days	53.50	10	5.350	4.330	425	
3 days	188.40	5	37.680	32.330	604	
4 days	597.50	5	119.500	81.820	217	
4½ to 5 days	[2] 2,374.70	[2] 15	[2] 158.313	[2] 38.813	[2] 32	Larvæ sealed.
Lot 3:						
1 day	5.70	9	0.633	[1] 0.530	[1] 530	
2 days	45.10	5	9.020	8.387	1,325	
3 days	114.80	5	22.960	13.940	155	
4 days	638.80	5	127.760	104.800	456	Sealing begun.
4½ to 5 days	[2] 2,374.70	[2] 15	[2] 158.313	[2] 30.553	[2] 24	Sealing completed.
Lot 4:						
1 day	3.05	5	0.610	[1] 0.510	[1] 510	
2 days	31.75	5	6.350	5.740	941	
3 days	166.50	5	33.300	26.950	424	
4 days	502.70	5	100.540	67.240	202	Sealing begun.
4½ to 5 days	[2] 2,374.70	[2] 15	[2] 158.313	[2] 57.773	[2] 57	Sealing completed.
Lot 5:						
1 day	5.20	10	0.520	[1] 0.410	[1] 420	
2 days	29.60	10	2.960	2.440	469	
3 days	70.10	5	14.020	11.060	374	
4 days	295.20	5	59.040	45.020	321	
5 days	727.80	5	145.700	86.660	147	Sealing begun.
5½ to 6 days	[2] 2,374.70	[2] 15	[2] 158.313	[2] 12.613	[2] 9	Sealing completed.
Average of Lots 1 to 5:						
0 day	1.00	10	0.100			
1 day	28.60	44	.650	0.550	550	
2 days	187.50	40	4.687	40.370	621	
3 days	616.10	25	24.640	19.928	426	
4 days	2,367.30	25	94.692	70.052	284	
4½ to 5 days	3,152.85	20	157.642	62.950	66	Larvæ attain maturity.

[1] Based on difference between the first day's weight and the weight of the egg as given for Lot 1.
[2] Figures obtained from weighing in 1916 of 15 mature larvæ from the same colony as Lot 1.

TABLE 2.—*Lot 8 (1922): The average weight in milligrams and the rate of growth of honeybee larvæ at different age periods in days from the time of hatching from the egg, under varying conditions.*

Age.	No nectar flow.			Stimulative feeding.			Effect of feeding.		Final average.	Rate of growth.		Table 1, Lots 1-5.		Straus.
	Date.	Weights.[1]	No.	Date.	Weights.[1]	No.	Difference in weights.	Per cent increase.		Average daily gain.	Per cent daily gain.	Weights.	Per cent daily gain.	Weights.
Egg												0.100		0.060
1 day [2]										[3] 0.550	[3] 550	.650	550	.300
2 days [2]	Aug. 11	3.96	5											
	Do	4.68	5											
	Do	5.30	5											
	Do	4.86	5											
	Do	5.34	5											
	Do	5.20	5											
	Do	3.80	5											
	Aug. 14	4.60	5											
	Do	4.48	5											
	Do	4.62	5											
	Do	4.70	5											
	Do	5.40	5											
Average		*4.745*							4.745	4.095	630	4.687	621	3.400
3 days [2]	Aug. 12	20.96	5											
	Do	24.12	5											
	Do	20.36	5											
	Do	18.96	5											
	Aug. 15	29.02	5											
	Do	21.16	5											
	Do	26.38	5											
	Do	29.52	5											
	Do	22.46	5											
	Do	27.28	5											
	Do	24.64	5											
	Do	29.66	5											
	Do	25.64	5											
	Do	25.10	5											
	Do	24.14	5											
Average		*24.626*							24.626	19.881	419	24.640	426	33.300

[1] Averages in italics. [2] Unsealed. [3] From Table 1.

TABLE 2.—*Lot 8 (1922): The average weight in milligrams and the rate of growth of honeybee larvæ at different age periods in days from the time of hatching from the egg, under varying conditions.*—Continued.

Age.	No nectar flow.			Stimulative feeding.			Effect of feeding.		Final average.	Rate of growth.		Table 1, Lots 1-5.		Straus.
	Date.	Weights.[1]	No.	Date.	Weights.[1]	No.	Difference in weights.	Per cent increase.		Average daily gain.	Per cent daily gain.	Weights.	Per cent daily gain.	Weights.
4 days [1]	Aug. 16	80.86	5	Aug. 30	94.96	5								
	Do.	90.48	5	do.	97.50	5								
	Do.	98.08	5	do.	93.38	5								
	Do.	84.44	5	do.	94.60	5								
	Do.	102.56	5	do.	94.40	5								
	Do.	100.62	5	do.										
	Do.	107.37	5											
	Do.	82.62	5											
Average		*93.378*			*94.968*		1.590	1.7	93.990	69.364	282	94.692	284	100.100
5 days	Aug. 14	[1] 142.38	5	Aug. 31	[2] 149.58	5								
	Do.	[1] 146.88	5	do.	[2] 155.40	5								
	Aug. 15	[1] 142.02	5	do.	[2] 151.58	5								
	Do.	[1] 146.82	5	do.	[2] 146.02	5								
	Do.	[1] 140.92	5	do.	[2] 148.00	5								
	Do.	[1] 142.62	5	do.	[2] 156.48	5								
	Do.	[1] 140.22	5											
	Aug. 17	[1] 142.88	5											
	Do.	[1] 142.52	5											
	Do.	[1] 151.98	5											
	Do.	[1] 149.32	5											
	Do.	[1] 141.54	5											
	Do.	[1] 149.92	5											
	Do.	[2] 147.88	5											
Average		*144.85*			*151.177*		6.327	4.4	146.748	52.758	56	157.642	66	134.500
6 days [3]	Aug. 15	152.54	5	Aug. 31	159.13	25								
	Do.	155.92	5	do.	164.99	25								
	Do.	152.52	5											
	Do.	155.70	5											
	Aug. 18	147.92	25											
	Do.	148.65	25											
Average		*149.966*			*162.06*		12.094	8.1	155.005	8.257	5.6	[4] 162.900	[4] 3	153.200

7 days[3]	Aug. 16	138.60	5	Sept. 9	148.22	5								
	Do	134.24	5	do	145.68	5								
	Do	132.30	5	do	148.04	5								
	Do	143.90	5											
	Do	144.36	5											
	Do	139.49	5											175.600
Average		*138.815*			*147.31*		8.495	6.1	141.647	−13.357	−8.6	[4] 147.700	[4] −9.0	147.200
8 days[3]	Aug. 17	138.92	5	Sept. 8	144.76	5								
	Do	130.26	5	do	142.92	5								
	Do	135.54	5	do	143.70	5								
	Do	136.98	5											
	Do	133.20	5											
	Do	133.68	5											
	Do	136.17	5											
	Do	134.76	5											
	Do	135.10	5											
Average		*134.956*			*143.79*		8.834	6.5	137.165	−4.482	−3.2	[4] 141.200	[4] −4.4	143.000
9 days[3]	Aug. 18	133.62	5											
	Do	130.76	5											
	Do	133.16	5											
	Do	134.56	5											
	Do	133.66	5											
Average		*133.152*							133.152	−4.013	−2.9			142 000

[1] Unsealed. [2] From Table 1. [3] Sealed. [4] From manuscript notes, Lots 1 to 7.

TABLE 3.—*Ratio of weight, in milligrams, of unconsumed food in cell, to weight of larva at different approximate age periods.*

Approximate age.	Number of observations.	Average weight of food per cell.	Average weight of larvæ.	Per cent ratio, food to larva.
Egg	Several.	None.	0.10	0
1.1 days	33	3.96	1.02	388
1.4 days	131	3.23	1.36	238
2.1 days	24	9.10	7.20	126
2.6 days	35	11.79	17.48	67
3 days	25	5.05	25.22	20
3.5 days	65	7.75	63.46	12
4.4 days	17	8.76	115.15	8

TABLE 4.—*Composition of larval foods—von Planta* (7).

Composition of dried substance.	Queen.	Drones.		Workers.	
		Under 4 days.	Over 4 days.	Under 4 days.	Over 4 days.
	Per cent.	*Per cent.*	*Per cent.*	*Per cent.*	*Per cent.*
Nitrogenous products	45.15	55.91	31.67	53.38	27.87
Fat	13.55	11.99	4.74	8.38	3.69
Sugar	20.39	9.57	38.49	18.09	44.93

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PART II. THE FEEDING OF HONEYBEE LARVÆ.

CONTENTS.

INTRODUCTION.

The larvæ of most bees are supplied with a mass of food consisting of a mixture of pollen and honey, which is put in place before the egg is laid. The larvæ of the honeybee, on the other hand, are fed at frequent intervals during the period of larval life. This behavior of the honeybee involves the services of nurse bees in caring for the larvæ. The nurse bees are always worker bees, usually those less than a month old. When worker bees are acting as nurse bees they take honey (or nectar) and pollen from adjacent cells of the comb and from these two materials elaborate the food on which the larvæ live and grow to maturity.

The food given to the youngest larvæ is a grayish-white paste-like material which is placed in the cell beside the larva. For older larvæ the prepared food is mostly fed directly to the larvæ. To supply food to a larva the nurse bee must enter the cell head first, so that it is impossible for one to see what occurs in the cell while the nurse bee is so situated. Because of the fact that the larva is wholly hidden while the worker is attending it, one can determine only the total time spent in this work, without being able to divide the time according to the various actions which may occupy the nurse bee during this activity. Attention was attracted to this phase of the feeding problem by the observation that nurse bees spend considerably more time in cells containing well-developed larvæ than they do in cells containing eggs or young larvæ. To get accurate data on the inequality of the time spent in feeding the present observations were begun.

As explained in Part I of this bulletin, Mr. Sturtevant was at the same time engaged in determining the rate of growth of the worker larvæ. His results showed such a striking correlation with those obtained for the time spent by the nurse bees in feeding that it was decided to cooperate in determining the correlation of the weight of larval food contained within the cell during each day of the larval period with the weights of the larvæ for each day. These results have been incorporated in Part I.

METHODS.

All observations were made on a colony of bees in a dark room, using artificial light. The hive used was especially designed to exclude all light from the brood nest except that used by the observer.

An entrance to the hive was gained by means of a maze tunnel leading through the wall of the room which excluded all light. The bees traveled in total darkness for a distance of approximately 6 inches before entering the hive proper.

The reason for placing this hive in a dark room was that when bees are kept in an observation hive out of doors they tend to congregate on the glass and thus hide the activities of the nurse bees to too great a degree for such observations. It was found that this does not occur when bees are kept in such a hive in a dark room for a considerable period.

The hive used was of standard 10-frame dimensions, containing 10 drawn combs which previously had been used for rearing brood. Several of these combs contained brood when placed in the hive and some of the combs contained sealed honey above the brood. The colony was of average strength for a colony kept in a single hive body. During the course of the observations the worker bees apparently flew in normal numbers to the field. Supplies were collected and brood was reared in numbers that justify the statement that the behavior of the bees, both workers and queen, was approximately normal. The observations extended from August 25 to September 9, 1922, during which period little nectar was coming in. A small amount of sirup was given once.

Various arrangements of electric lamps were tried so as to secure the best lighting, but best results are obtained by using a 40-watt lamp, which is placed on the top of the hive and back from the edge far enough to exclude all direct rays from the comb. A mirror attached to a flexible support is then adjusted so that the rays from the lamp are reflected to the portion of the comb under observation. The hive is slightly above the level of the eyes, and when the eyes are placed just behind and a little below the mirror an excellent view of the interior of the cell is obtained. A concave mirror is best for close inspection of the contents of a cell, while a small plane mirror is best for ordinary observations. Shades are attached to the sides of the mirror to protect the eyes from direct light from the lamp.

Under normal conditions in a hive containing a number of combs the brood nest is in the inner combs, and this makes impossible observations of what takes place during feeding. To obviate this difficulty, combs containing larvæ of the desired age were taken from the center of the brood nest and placed next the glass. The hive was well filled with bees of all ages, and at once the nurse bees appeared and took up their duties as attendants on the outside of the comb, where their activities could be observed readily.

On one occasion all brood was removed from the hive and empty combs partially filled with sirup were put in. A single frame of brood of the desired age was then placed in the hive next the glass. On this occasion the queen appeared on the outside of this comb, where she remained for one hour. During this period she laid a number of eggs, rested, and was fed, in a manner suggesting normal conditions. Several flashlight pictures of the laying queen were taken, but even these flashes failed to drive her to the other side of the comb.

The cells to be studied are either marked before the comb is placed in the hive or selected by a close study with the concave mirror after

the comb is placed in the hive. These cells are then marked by pasting a small triangular pointer on the outside of the glass, the fine point of which comes directly over the cell to be studied. These pointers serve to hold abbreviated data concerning the particular cell which they mark; they interfere in no wise with the observations and are permanent. Paint or marks on the edge of the cell itself are soon removed, and the bees working at this removal may introduce a source of error.

Preliminary observations were made, and it was decided that 10-minute periods for observations are probably of the most practical length. Such a short period can be used to obtain data on a greater number of different cells within a given time, or they can be doubled and several readings taken in succession on the same cell, in case it appears that either procedure is desirable.

NURSE BEES.

A nurse bee moves about over the brood combs when attending to the needs of eggs or larvæ. She stops sometimes and lowers her head over certain cells, and at other times she thrusts her head within the brood cells but promptly withdraws it. On still other occasions she enters a cell containing an egg or a larva and remains in it for a more or less extended period, presumably engaged in certain activities. The activities of the nurse bees in their work may be classified according to the time spent at each cell and according to the probable purpose of their visits. These actions are here divided into inspections and nursing, and the inspections are further subdivided.

INSPECTION, TYPE A.

A nurse bee in pursuing her duties sometimes pauses with her head over an open brood cell. When she does this she usually lowers her head slightly, barely perceptibly at times, and then passes on. It is observable that she is a nurse bee, and presumably this behavior is part of the activity connected with her duties as a nurse, for she has just come from another cell where she has been feeding a larva; and if her movements are followed, she is seen to enter another cell, where her movements indicate that she is attending to the wants of the inmate.

This act of pausing, together with other activities which may accompany it but which have not as yet been explained, is difficult to classify, because its significance is not known. Such a pause may be all that is required to gain information through some sense organ concerning the feeding requirements of the inmate, and therefore this activity must be recorded. This type of activity is frequently noted, particularly when the cell contains an egg or a young larva.[1] At present certain and special significance can not be attributed to this activity other than calling attention to it as a fact; it was decided to enter it in the data under the designation Inspection, Type A.

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[1] A similar hesitating behavior is noted in the case of the laying queen, and it is seen most often when she is laying in a comb containing scattered brood and eggs. At such times she frequently pauses above certain cells and then passes on without entering. Many cells thus passed have been examined and practically without exception they contained either eggs, larvæ, or noticeable débris.

INSPECTION, TYPE B.

On other occasions a nurse bee approaches an open brood-cell, pauses, lowers her head and thrusts it into the cell, but withdraws it almost immediately. The whole head may be thrust within the cell and perhaps part of the thorax as well. When the whole time of this operation does not exceed two seconds, it is assumed that there is not time for the nurse bee to do anything in the nature of actual feeding. Two seconds might allow time for the ejection of a liquid food, but such a short time is probably insufficient for a determination of the already available food supply or the needs of the larva and then additional time for supplying such food as may be needed. In taking data on the time spent in caring for the brood, all such activities are designated Inspection, Type B. That bees do determine the quantity of food present is evidenced by the fact that all larvæ of approximately the same age and position on the comb have about the same amount of food at all times. This would not be the case if the different nurse bees did feeding indiscriminately and without a determination of the amount of food already within the cell. There is opportunity in the form of activity under this heading for tactual contact with the larva, while in the former type (Type A) there is no such opportunity.

NURSING.

Feeding of the larva occurs when the nurse bee enters the cell, but obviously an appreciable time is required for this activity. For such activity the descriptions under the two preceding types of inspection are not valid. While the nurse bee is within the cell, activities other than actual feeding may occupy part of her time, but probably all these activities have to do with the care of the larva.[2]

The time spent by a nurse bee within the cell engaged in feeding varies between 2 seconds and from 3 to 4 minutes. Even the shortest periods may be distinguished from inspection by the fact that there is a noticeable pause of the bee while within the cell. The longest periods mentioned are quite exceptional. It is impossible to see what a nurse bee is actually doing within a cell, but some basis for a differentiation between inspecting and nursing visits should be adopted, even though it appears more or less arbitrary. The most practical basis appears to be that of time. When the time spent within the cell is less than 2 seconds the visit is classed as an inspection, and when the visit is of longer duration it is considered under the designation nursing. This basis does not appear so arbitrary to the observer, since the actual behavior in these two cases appears rather more distinctive than the difference in time alone would cause one to believe. While it is quite possible that occasionally an inspecting bee may spend more than two seconds, it is nevertheless true that most inspection visits are completed in less than two seconds.

Having, therefore, decided upon this method of classification of the behavior of the nurse bees, observations were begun. The results

[2] Occasionally a worker bee creeps into a brood-cell containing a small larva merely to rest. Such a worker may usually be distinguished promptly from a nurse bee engaged in her duties of feeding.

obtained in the observations are presented in Tables 5 to 7 and in Figures 12 and 13.

NUMBER OF VISITS.

Observations were made for each group of larvæ to determine the actual number of visits of nurse bees to the eggs and larvæ under observation, each egg or larva being watched individually for a 10-minute period, as previously stated. The data given in Tables 5 to 7 show the details of these observations. The average number of visits for a 10-minute period to the egg and to larvæ of ages from 1 day to 5 days were, respectively, as follows: 5.93, 6.40, 5.79, 8.08, 14.47, 19.83 (Table 6 and fig. 12). Of the visits listed as Inspection, Type A, there is no great change in the number as the larvæ increase in size and age, the slight variations being probably without great significance (Table 5). The number of visits listed as Inspection, Type B, increases as the larvæ grow larger and older, while the number of visits listed as nursing, in which the nurse bee stays in the cell at least two seconds, show no great significant variation until the fourth day of larval life, when there is a great increase in the number, followed by still further increase on the fifth day. It is significant that there is no marked decrease in the total number of visits on the second day of larval life, although, as will be discussed later, there is at this time a decrease in the amount of nursing.

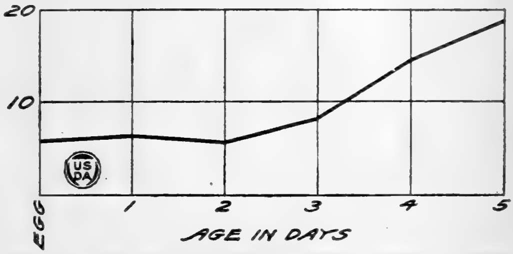

FIG. 12.—Showing the relation of the number of visits made in nursing honeybee larvæ per 10-minute period to age. (Table 6.)

From the foregoing it readily can be seen that the egg and larva are under the almost constant observation of the nurse bees, and that under such a system of visitations there is little likelihood of any individual being overlooked for any considerable length of time.

TIME SPENT IN NURSING.

Observations were also made for each age to determine the total time spent by the nurse bees with the eggs and larvæ under observation, each egg or larva being also watched for a 10-minute period. The details are given in Table 5. The average time in seconds spent by nurse bees for a 10-minute period with the eggs and larvæ of ages from 1 to 5 days were, respectively, as follows: 6.5, 20.73, 5.93, 11.42, 41, 118.08 (Table 7 and fig. 13). It should be noted that the visits here discussed are only those listed as nursing, and no account is here taken of the shorter visits listed as inspection of the two types. There is observed a decided increase in the amount of time spent in nursing as the larvæ grow in size and age, the outstanding exception being for larvæ 2 days of age, which will be discussed later. The total time spent in feeding during the five days of larval life is 6.57 per cent of the whole time, while on the fifth day, when the larvæ are largest and require the most food for their development, the

nurse bees actually spend within the cell 19.68 per cent of the whole time. These figures indicate the unceasing care of the developing bees even more than do the data on the number of separate visits made to them by the nurse bees.

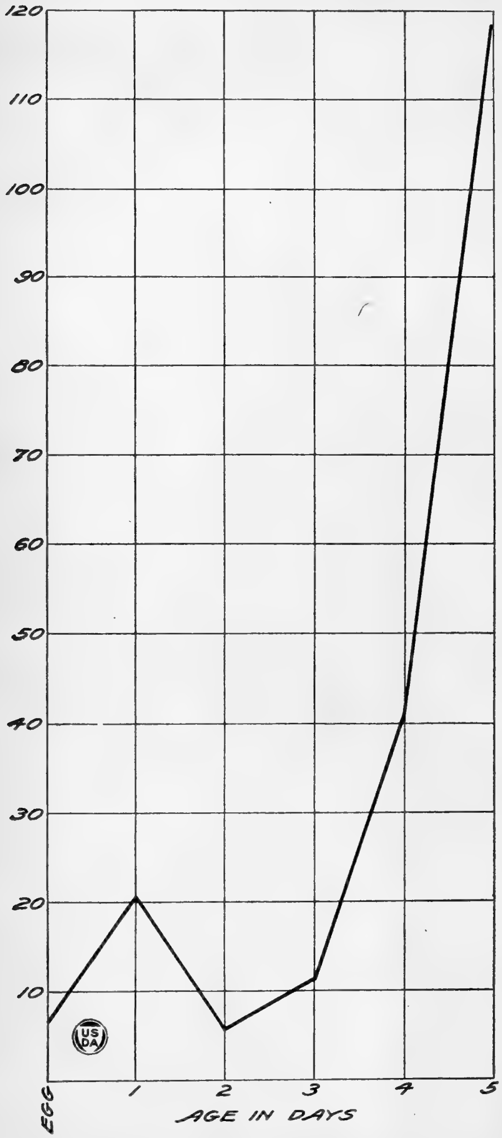

FIG. 13.—Showing the relation of average time (seconds) spent in nursing honeybee larvæ per 10-minute period to age. (Table 7.)

Few successive observations on individual larvæ were made, since the number of visits and the time spent within the cells were fairly uniform for each age. Occasionally, where a particularly small or a particularly large total was obtained for a certain cell in one 10-minute period, a second observation was made at once. Usually in such cases the average of two such readings was about equal to the average per 10-minute period for the given age. It therefore appears that if a cell has received more than the usual amount of attention during one observation period of 10 minutes, the nurses during the ensuing period devote less than the average amount of attention to that particular cell, though they may inspect it as many or even more times than they did during the preceding observation period. Likewise any slighting of a certain cell during one period is compensated in a subsequent period by more than the average amount of attention.

CHANGE IN FEEDING BEHAVIOR.

It has long been known that the developing larva is not fed the same kind of material throughout larval life, it being usually believed that the change in the character of the food occurs on the fourth day. The data herein recorded show not only that such

a change does occur, but that there is also a remarkable change in the type of feeding, and that this change occurs at the time when the character of the food changes. It thus becomes necessary to point out that the change in food occurs not on the fourth day, as commonly stated, but much earlier. This is further substantiated by the work done by Sturtevant and the writer, recorded in Part I of this bulletin.

As has been stated earlier, the number of visits for each 10-minute period averages 5.93, 6.4 and 5.79 for the egg, 1-day, and 2-day larvæ, respectively. For these same ages the time in seconds spent in nursing (each period more than two seconds) is as follows: 6.5, 20.73, and 5.93. Such a change, on superficial consideration, appears inconsistent, since one naturally supposes that a 2-day larva would require more visits and more time spent in nursing than does the egg or the 1-day larva. The time spent in nursing the 1-day larva is actually over three times as great as the time for the 2-day larva.

This whole matter is unexplained until it is viewed in the light of the results obtained by Sturtevant and the writer in Part I of this bulletin on the amount of food found within the cell for larvæ of different ages. No food is ever found in the cell with the egg, while the weight of food within the cells containing 1-day and 1.4-day larvæ averages 3.96 and 3.23 milligrams, respectively. If now these findings are taken into consideration with the well-established fact that there is a change in the character of the food of the worker larva, the whole matter is explained. Immediately following the hatching of the egg, the nurse bees surround the young larva with food in relatively large quantity. As pointed out, this first supply of food may weigh 3.96 milligrams per cell, which is equal to almost four times the weight of the young larva which it supplies. These figures indicate that, as already suggested, a large amount of food is placed in the cell of the recently hatched larva and that during the second day little or no feeding is done, since the attention given (in number of visits as well as in time in nursing) is about equal to that given the egg. Since the egg is never surrounded by food, this indicates that the attention given the 2-day larva is of no greater significance than that given the egg, so far as the giving of food is concerned. That there is a rapid giving of food on the first day followed by little or no supplying of food on the second day is further supported by a comparison of the weights of food contained in the cells with larvæ of these ages. Nelson and Sturtevant show that there is a decrease of 0.73 milligram in the amount of food in the cell on the second day. During this interval there is an increase in the weight of the larva of 0.34 milligram so that the decrease in weight of available food exceeds the increase in weight of the larva. The difference (0.39 milligram) is doubtless accounted for by the total combustion of food by the rapidly growing larva. Larvæ of this age are doubtless too small to ingest more than the amount of food represented by the decrease in weight of the available food, from which it follows that little or no food is given the larva on the second day.

On the third day there is a slight increase in the amount of time spent by the nurse bees within the cell, and this indicates that feed-

ing is being resumed. Further evidence to substantiate this conclusion is found both by weighings of the food present and by a comparison of the composition of food found in cells containing 2-day and 3-day larvæ. Sturtevant shows that there is a considerable increase in the amount of food found in cells containing 3-day larvæ over the amount found with 2-day larvæ. The composition of the food is changed also by the third day. This is most conspicuous through the fact that the color of the food found within the cells containing larvæ of 3 days or more varies with the color of the pollen being brought in from the field, no stores of old pollen being present in the hive used in this experiment. The larval food found in cells containing 1-day and 2-day larvæ is of a uniformly grayish-white color.

With the change of food it appears that a different method of feeding is gradually adopted, mass feeding giving place to progressive feeding; that is, the food is no longer given in quantity and in excess of immediate needs, but is supplied at about the same rate at which it is ingested. The bee larva is a rapidly growing organism, hence it follows that if the food is supplied from time to time, it must be introduced in rapidly increasing amounts. This necessitates either more visits or longer feeding periods, or both of these factors may be increased. The last is actually the case. The number of visits is increased, particularly on the fourth and fifth days, and the time of the visits is notably lengthened.

Mass feeding, if this is strictly what happens in the case of the food first supplied, taken together with the later progressive feeding, shows an interesting similarity to the behavior of certain solitary bees, some of which practice mass feeding exclusively, while others, recently investigated, practice progressive feeding. It also has a resemblance to the feeding behavior of the bumblebee, which lays the eggs on a lump of "pollen-paste" to furnish the first food after hatching, and then some time after the larva is hatched a type of progressive feeding is begun. A similarly modified type of mass feeding is shown in the case of the queen larvæ of the honeybee. The large queencell is rapidly crowded with food before it is sealed, and the larva completes its development while eating more food which is inclosed with it. Frequently a mass of dried food remains even after the emergence of the queen, which confirms the supposition of mass feeding in this instance.

The fact that the food of the larva up to 2 days is placed in the cell beside the larva and is apparently not fed into the mouth seems to preclude the possibility of mutual feeding of the nurse bee and larva during this period. Such reciprocal feeding has been described for certain social wasps and ants, and to this phenomenon Wheeler has given the name trophallaxis. Since the queen larva is also fed by mass feeding, perhaps exclusively by that method, there is little or no indication of trophallaxis in this instance. Roubaud has shown that in certain wasps in which trophallaxis is observed there is a decided disproportion between the amount of material given by the nurse wasp and that received by it from the larva being fed, the secretion received being sometimes greater than the food given by the nurse. Reciprocal feeding has never been observed in the honeybee, but since the food given to the older larvæ is not complex in character and probably requires no great amount of work on

the part of the nurse bees for its elaboration, it is difficult to explain the great amount of time spent by the nurse bees in the cells merely on the basis of the belief that food is being fed to the larva all that time. That the usual method of feeding is directly to the larval mouth is clear from the fact that no significant amount of food is ever found in the base or on the sides of the cell of an older larva. In the determinations of the weight of food residue in the cell it is noteworthy that for each age it is quite uniform. If one dared to assume reciprocal feeding, this might perhaps account for the peculiar development of the worker bee in the repression of the sex organs and other morphological modifications. Wheeler has shown that parasites like Orasema, by the withdrawal of food substance already assimilated by ant larvæ, may bring about morphological changes of the same kind as those which distinguish the worker ant from the queen.

The number of inspections and visits to the egg may be accounted for, in the light of present knowledge, only by the assumption that they are due to the watchfulness of the nurses, since they feed the young larvæ promptly on hatching. In no other cases except in the ants are insects known to care for the eggs. Ants lick the eggs, but this is supposed to be for the purpose of sticking them together so that they may be transported in quantity.

THE LABOR OF THE NURSE BEES.

Contrary to the common belief, it was noted that bees are not outstanding examples of industry. During these observations much resting was observed both among the nurse bees and among the other workers. Demuth has observed that the average number of trips to the field often does not exceed four per day, whereas the time required for making such trips and the time required to deposit the supplies obtained might readily permit a much larger number. Such an apparent lack of industry concerns the present problem only in that it is evident that more nurses will be required for a given number of larvæ in proportion to the time that is taken off from their active duties. In the case of nurse bees, however, other considerations enter, since considerable time may be required for the elaboration of larval food. Thus it is quite possible that a nurse bee, though apparently idle so far as one can determine, may be active physiologically in the production of larval food. Any determination of the efficiency of a single nurse bee may be made only by observations on a large number of individual nurse bees extending throughout the day. Since the time actually spent in the cell is so great and since the time occupied in inspection and in passing from cell to cell is considerable, in the absence of detailed investigation of the efficiency of the individual nurse bee one may only conclude that a single nurse bee is able to care for but a few larvæ.

In the case of the bumblebee the queen is able to rear 6 to 16 larvæ, varying somewhat with the species. The ant queen rears 12 to 15 larvæ, but in this case, as in the case of the bumblebee, the first brood is composed of quite small individuals. Those of the bumblebee are often not much larger than house flies. All worker honeybees are approximately of the same size, and in case supplies are so low that it is impossible to feed all the larvæ a sufficient amount of food to

bring them to normal size, some of the eggs, or even some of the larvæ are removed so that the remaining ones may be sufficiently fed.

To gain some idea of the immensity of the task of rearing a single larva it is necessary to refer to Table 6. Averaging the results found for the eggs and larvæ of all ages, it is seen that on an average more than 1,300 visits are made in 24 hours. On the last day before capping no less than 2,855 visits are made by the nurse bees to a single cell. On this last day before capping approximately $4\frac{3}{4}$ hours are spent by the nurse bees within the cell.

When these facts are considered, together with the time required for obtaining, preparing, and transporting the food, and the time required for capping the cells, it is evident that the rearing of brood constitutes a considerable burden to the working forces of the colony, doubtless reducing the field work and also other inside labors. It follows, therefore, that any considerable brood-rearing during the main honey flow must reduce materially the surplus honey procured. Not only is the surplus honey cut down because of the reduction in the number of field workers and comb builders, but honey which is brought in is lost, since it is fed to larvæ which usually develop too late to aid in the gathering of a crop.

The foregoing statements are substantiated in practical beekeeping by the well-established fact that a newly hived swarm gathers honey and builds comb more rapidly than an equally strong colony which has not swarmed. The reason for this is now clear. All members of a swarm may be either field workers or comb builders, while at first none of the nectar brought from the field is fed to brood, all of it being stored or consumed by adult bees. Similar results are obtained by confining the queen to a single brood chamber just before the honey-flow, since this somewhat limits the queen in egg-laying and reduces the amount of brood to be cared for during the honey-flow. Caging the queen has a similar effect, although this introduces other factors which may reduce the amount of surplus honey that might be obtained under this system.[3]

SUMMARY.

The attention given eggs and larvæ of the honeybee by the nurse bees consists in visits to the cells for purposes of inspection and for work or nursing carried on within the cell.

The number of such visits averages about 1,300 per day during the eight days from the time the egg is laid till the fully grown larva is sealed within the cell.

The elaborated food on which larvæ feed during about the first two days of the larval period is practically all placed in the cell with the newly-hatched larva soon after hatching. This is mass feeding.

Soon after the second day another kind of food is supplied to the worker larva. This food contains considerable undigested pollen and is fed at approximately the same rate at which it is consumed by the larva.

The fact that the method of feeding the worker larva is changed, for no apparent reason, after the second day, together with the excessive amount of time spent in visits of nurse bees to the older worker larvæ, suggests that there may be here a reciprocal feeding

[3] Phillips, E. F. Beekeeping. A discussion of the honeybee and of the production of honey, 457 p., 190 fig., front. New York, 1915. [See p. 282.]

between the nurse bees and the older worker larvæ, as has been observed in related insects, such as ants and wasps.

When a certain larva has received more than the average amount of nursing for larvæ of its age during one observation period, less than the average amount of nursing is given it during the following period. A conspicuous lack of nursing in one period is usually followed by more than average nursing in a subsequent period.

Over 10,000 visits are made to each developed individual bee during the eight days from the time the egg is laid until the cell is capped. During this same period a considerable amount of time is spent by the nurse bees actually within the cell. The time thus spent during the last day before capping is nearly 4¾ hours. A consideration of these facts, together with the time required for procuring, elaborating, and transporting the food to the cells, and also the time required for capping the cells, leads one to conclude that the number of bees engaged in nursing must be very large.

Beekeepers have recognized the great cost in honey and labor of the rearing of brood. They have, of course, recognized the desirability of rearing large amounts of brood before the honey-flow, but they also recognize the desirability of a reduction in brood-rearing after the honey-flow begins. They have to some extent accomplished the latter object by the removal of brood, by the caging of the queen, or by the reduction of the space available for egg-laying by the queen, these manipulations also accomplishing other purposes needful in certain phases of beekeeping (swarm control, comb-honey production, etc.). While these manipulations serve the purposes for which they are intended, they often introduce other factors which act adversely on the nectar gathering of the colony. One may not therefore from single facts regarding the excessive labor cost of brood-rearing recommend any special manipulations of the colony for the purpose of increasing the total honey crop, without a study of all the factors involved.

TABLE 5.—*Data obtained from observations of visits of nurse bees to cells containing eggs and larvæ.*

VISITS TO CELLS CONTAINING EGGS.

Designation.	Number of visits per period of 10 minutes.				Time spent in nursing[1] (seconds).	
	Inspection, Type A.	Inspection, Type B.	Nursing.[1]	Total.	Time spent in individual visits.	Total time spent per 10-minute period.
					Seconds.	*Seconds.*
Egg a	2	3	1	6	2	2
Egg b	3	4	0	7	0	0
Egg c	0	2	2	4	3, 19	22
Egg d	2	10	4	16	2, 3, 8, 4	17
Egg e	7	0	0	7	0	0
Egg f	5	0	1	6	5	5
Egg g	0	3	0	3	0	0
Egg h	1	2	0	3	0	0
Egg i	0	3	1	4	9	9
Egg j	0	3	3	6	4, 8, 2	14
Egg k	0	1	3	4	4, 4, 2	10
Egg l	1	2	2	5	2, 8	10
Egg m	1	3	0	4	0	0
Egg n	1	6	1	8	2	2
Average	1.64	3.0	1.29	5.93	5.06	6.50

[1] Under this heading are given visits of two seconds or more. There is no actual giving of food to the egg.

TABLE 5.—*Data obtained from observations of visits of nurse bees, etc.*—Contd.

VISITS TO CELLS CONTAINING LARVÆ ONE DAY AFTER HATCHING.

Designation.	Number of visits per period of 10 minutes.				Time spent in nursing (seconds).	
	Inspection, Type A.	Inspection, Type B.	Nursing.	Total.	Time spent in individual visits.	Total time spent per 10-minute period.
					Seconds.	*Seconds.*
Larva a	0	5	2	7	14, 29	43
Larva b	2	5	0	7	0	0
Larva c	3	4	1	8	2	2
Larva d	0	2	3	5	4, 20, 4	28
Larva e	2	3	3	8	2, 4, 4	10
Larva f	2	1	3	6	18, 8, 11	37
Larva g	0	8	2	10	2, 2	4
Larva h	0	4	2	6	13, 70	83
Larva i	0	0	1	1	2	2
Larva j	0	6	3	9	4, 2, 4	10
Larva k	0	6	0	6	0	0
Larva l	0	3	1	4	7	7
Larva m	0	3	1	4	60	60
Larva n	2	5	1	8	3	3
Larva o	0	4	3	7	7, 3, 12	22
Average	0.73	3.93	1.74	6.40	12.00	20.73

VISITS TO CELLS CONTAINING LARVÆ TWO DAYS AFTER HATCHING.

Larva a	0	5	1	6	2	2
Larva b	1	5	1	7	4	4
Larva c	1	3	2	6	3, 3	6
Larva d	1	2	3	6	3, 5, 2	10
Larva e	0	1	1	2	2	2
Larva f	0	5	0	5	0	0
Larva g	0	6	1	7	5	5
Larva h	0	4	2	6	2, 2	4
Larva i	0	3	0	3	0	0
Larva j	0	3	1	4	5	5
Larva k	2	0	1	3	2	2
Larva l	1	8	5	14	4, 8, 4, 12, 3	31
Larva m	0	4	3	7	2, 3, 4	9
Larva n	1	3	1	5	3	3
Average	0.50	3.71	1.57	5.79	3.77	5.93

VISITS TO CELLS CONTAINING LARVÆ THREE DAYS AFTER HATCHING.

Larva a	5	10	3	18	2, 5, 3	10
Larva b	7	5	0	12	0	0
Larva c	1	8	6	15	2, 3, 4, 3, 2, 4	18
Larva d	0	7	0	7	0	0
Larva e	0	6	2	8	36, 3	39
Larva f	0	3	0	3	0	0
Larva g	2	2	2	6	6, 8	14
Larva h	0	2	1	3	2	2
Larva i	2	3	1	6	3	3
Larva j	2	3	0	5	0	0
Larva k	0	4	4	8	7, 2, 2, 33	44
Larva l	1	4	1	6	7	7
Average	1.67	4.75	1.67	8.08	6.85	11.42

VISITS TO CELLS CONTAINING LARVÆ FOUR DAYS AFTER HATCHING.

Larva a	3	6	10	19	4,3,2,5,3,2,3, 2,3,30.	57
Larva b	5	17	10	32	4,2,4,4,2,2,2, 4,2,3.	29
Larva c	6	9	3	18	8,5,2	15
Larva d	3	3	5	11	13,2,2,3,5	25
Larva e	0	4	8	12	10,7,8,8,3,5, 23,8.	72
Larva f	0	6	1	7	3	3
Larva g	0	13	4	17	13,12,2,3	30
Larva h	0	8	5	13	2,13,2,3,13	33
Larva i	0	7	7	14	2,42,3,4,8,2,4	65
Larva j	1	10	4	15	27,2,8,30	67
Larva k	0	5	1	6	3	3

TABLE 5.—*Data obtained from observations of visits of nurse bees, etc.*—Contd.

VISITS TO CELLS CONTAINING LARVÆ FOUR DAYS AFTER HATCHING—Contd.

Designation.	Number of visits per period of 10 minutes.				Time spent in nursing (seconds).	
	Inspection, Type A.	Inspection, Type B.	Nursing.	Total.	Time spent in individual visits.	Total time spent per 10-minute period.
					Seconds.	*Seconds.*
Larva l	0	4	5	9	5, 2, 4, 2, 24	37
Larva m	0	8	8	16	3, 4, 2, 5, 4, 2, 6, 62.	88
Larva n	0	5	7	12	20, 5, 3, 2, 5, 11, 2	48
Larva o	0	11	5	16	5, 4, 25, 7, 2	43
Average	1.20	7.73	5.54	14.47	7.41	41

VISITS TO CELLS CONTAINING LARVÆ FIVE DAYS AFTER HATCHING.

Designation.	Inspection, Type A.	Inspection, Type B.	Nursing.	Total.	Time spent in individual visits.	Total time spent per 10-minute period.
Larva a	2	12	15	29	10, 2, 4, 3, 18, 3, 7, 18, 4, 11, 3, 3, 3, 2, 4.	95
Larva b	3	7	17	27	3, 25, 8, 8, 2, 7, 8, 7, 9, 2, 2, 20, 40, 2, 3, 6, 2.	154
Larva c	2	9	8	19	2, 2, 2, 5, 3, 2, 2, 2.	20
Larva d	0	14	6	20	58, 62, 14, 4, 110, 2.	250
Larva e	0	8	3	11	2, 2, 6	10
Larva f	0	16	7	23	2, 3, 2, 63, 2, 7, 3	82
Larva g	0	12	2	14	60, 10	70
Larva h	0	7	14	21	17, 23, 5, 15, 10, 70, 2, 7, 25, 45, 7, 15, 35, 15.	291
Larva i	0	11	10	21	60, 3, 2, 2, 4, 5, 6, 5, 7, 16.	110
Larva j	0	9	10	19	7, 2, 4, 3, 6, 3, 3, 3, 2, 2.	35
Larva k	0	10	5	15	3, 10, 9, 2, 45	69
Larva l	0	8	11	19	7, 15, 35, 3, 7, 40, 35, 35, 15, 15, 24.	231
Average	0.58	10.25	9.0	19.83	13.12	118.08

TABLE 6.—*Summary (from Table 5) of the number of visits of all kinds, each record being the total for an individual egg or larva for a 10-minute period.*

Age.	Number of visits of all kinds by nurse bees.															Average per period.
Egg	6	7	4	16	7	6	3	3	4	6	4	5	4	8	....	5.93
1-day larva	7	7	8	5	8	6	10	6	1	9	6	4	4	8	7	6.40
2-day larva	6	7	6	6	2	5	7	6	3	4	3	14	7	5	....	5.79
3-day larva	18	12	15	7	8	3	6	3	6	5	8	6	....	....	....	8.08
4-day larva	19	32	18	11	12	7	17	13	14	15	6	9	16	12	16	14.47
5-day larva	29	27	19	20	11	23	14	21	21	19	15	19	....	....	....	19.83

TABLE 7.—*Summary (from Table 5) of the time spent in nursing, each record being the total for an individual egg or larva for a 10-minute period.*

Age.	Time spent in nursing (seconds).															Average per period.	Average per visit.
Egg	2	0	22	17	0	5	0	0	9	14	10	10	0	2	....	6.50	5.06
1-day larva	43	0	2	28	10	37	4	83	2	10	0	7	60	3	22	20.73	12.00
2-day larva	2	4	6	10	2	0	5	4	0	5	2	31	9	3	....	5.93	3.77
3-day larva	10	0	18	0	39	0	14	2	3	0	44	7	....	....	....	11.42	6.85
4-day larva	57	29	15	25	72	3	30	33	65	67	3	37	88	48	43	41.00	7.41
5-day larva	95	154	20	250	10	82	70	291	110	35	69	231	....	....	....	118.08	13.12

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February 18, 1924.

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